

RHODODENDRON ALBIFLORUM HOOK.
(ERICACEAE): ONE TAXON OR TWO?

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ABSTRACT

Populations of *Rhododendron albiflorum* Hook. in Colorado are disjunct by ca. 1470 km from populations in the Pacific Northwest. Morphological data showed that plants from the two sets of populations differed in length and width of petals and length of stamens. Genetic identity figures based on analysis of isozyme electrophoresis found Colorado and Pacific Northwest populations to have an \bar{I} value of .76. These morphological, geographical and isozymic data support recognition of the Colorado plants as *Rhododendron albiflorum* Hook. var. *warrenii* (A. Nelson) M. A. Lane, *comb. & stat. nov.*; the Pacific Northwest plants are *Rhododendron albiflorum* Hook. var. *albiflorum*.

Key Words: *Rhododendron albiflorum*, morphology, isozyme electrophoresis, disjunct populations, Colorado, Pacific Northwest.

INTRODUCTION

Rhododendron albiflorum Hook. ("Cascades Azalea" or "White Rhododendron") is a small shrub of subalpine areas in the Cascade Mountains of northern California and Oregon, the Olympic Mountains of Washington, and the Rocky Mountains of British Columbia, Alberta and Montana, with disjunct (by ca. 1470 km) populations in the Rocky Mountains of Routt and Jackson Counties, Colorado (Figure 1). The plants have grayish bark and short-petiolate, narrowly elliptic, sparingly pubescent leaves. White flowers are borne in small axillary clusters; corollas are broadly campanulate and up to 2 cm wide. Though the plants are very attractive, they are extremely difficult to cultivate, and so the species is not well known among azalea fanciers.

Named by Hooker (1838), *Rhododendron albiflorum* is based on a specimen from "alpine woods of the Rocky Mountains" collected by Drummond, probably in Alberta or southeastern British Columbia during his journey there in 1825-27 (McKelvey, 1956). In 1900, Rydberg included Hooker's plant in *Rhododen-*

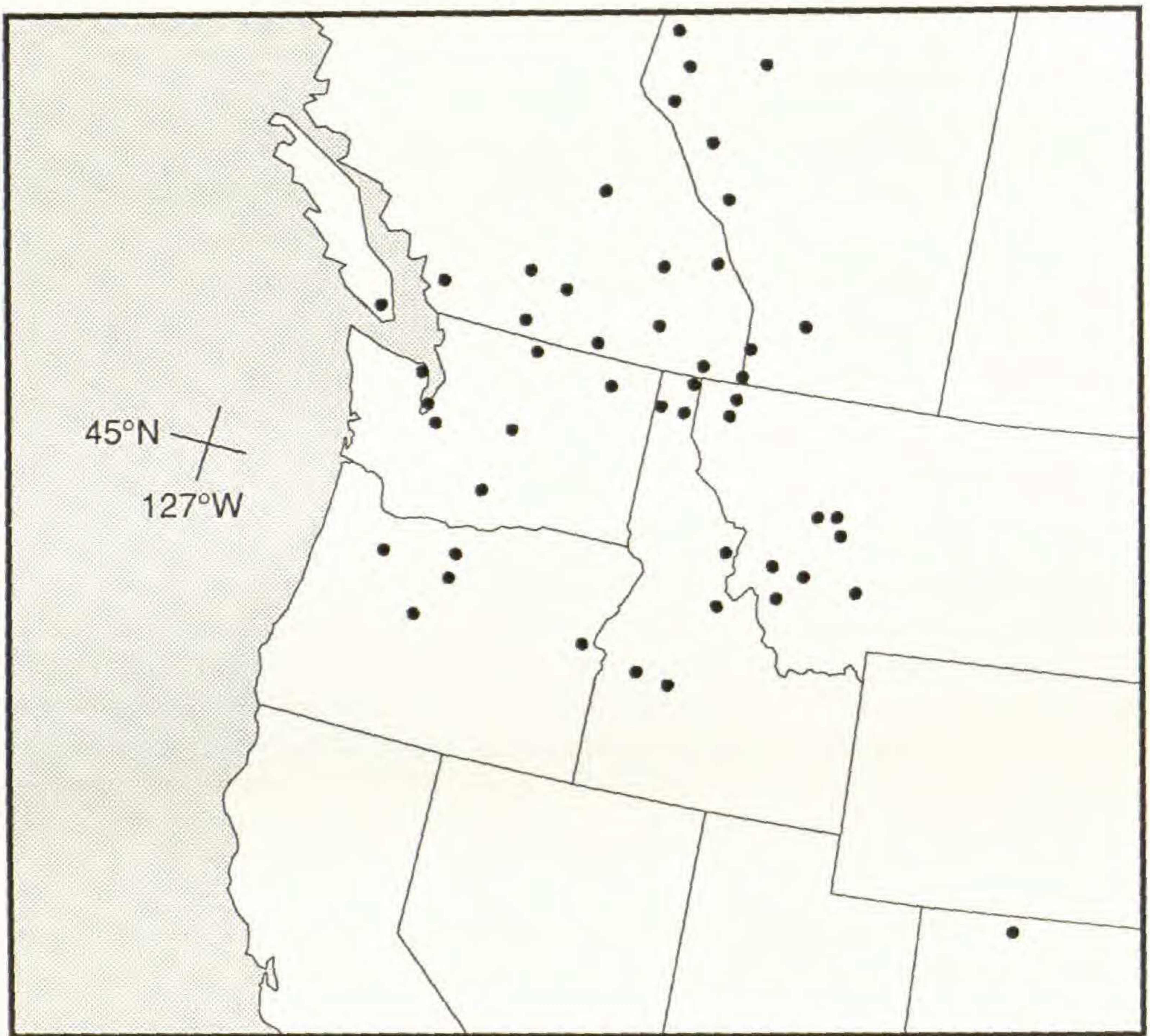


Figure 1. Distribution of *Rhododendron albiflorum* Hook. Each dot may represent one or more collections. The Colorado populations (Three Island Lake, Lake Diana, Lake Katherine, Lone Pine Creek, Gilpin Lake, Ute Pass and Slavonia) all lie within a 50 sq. mi. area in the Mount Zirkel Wilderness Area of Routt National Forest in Routt and Jackson Counties, Colorado.

dron subg. *Azaleastrum*, and elevated the subgenus to generic status. He contended that the deciduous leaves of *Azaleastrum albiflorum* (Hook.) Rydberg distinguished it from *Rhododendron*, and that its large sepals, lateral flowers, peltate stigmas and campanulate corollas separated it from *Rhodora* and *Azalea*. Nelson (1913) accepted Rydberg's generic treatment of the Pacific Northwest plants, but considered the Colorado populations sufficiently different to warrant specific status, and named them *Azaleastrum warrenii* A. Nelson. Most authors of floras (e.g., Harrington, 1954; Peck, 1961; Hitchcock and Cronquist, 1973) have accepted neither Rydberg's genus nor Nelson's species, though Weber (1990) and Weber and Wittmann (1992) use *A. albiflorum* (Hook.) Rydberg for the Colorado populations. A recent monographic treat-

ment (Philipson and Philipson, 1986) considered *R. albiflorum* to be the sole member of *Rhododendron* subg. *Candidastrum*, which nonetheless clusters with sect. *Azaleastrum* of subg. *Azaleastrum* in phenetic analyses (Palser et al., 1991).

In this study, we asked two questions: 1. Do the populations from the Pacific Northwest and those from Colorado constitute one taxon or two? 2. If there is more than one taxon present, should they both be recognized at the species level, or both as varieties of a single species? We addressed these questions using morphometric and isozyme electrophoretic techniques.

MATERIALS AND METHODS

Collection data obtained from a total of 712 specimens borrowed from 11 herbaria were used to construct a distribution map for the taxon (Figure 1). Pollen from a selection of these specimens was studied using both light and scanning electron microscopy. Eight quantitative characters, referred to below as variables five through twelve (VAR5 = leaf length, VAR6 = leaf width, VAR7 = number of flowers per cluster, VAR8 = sepal length, VAR9 = sepal width, VAR10 = petal length, VAR11 = petal width, and VAR12 = stamen length), were measured on representative specimens from the Pacific Northwest ($n = 51$) and from Colorado ($n = 7$). Variables one through four were geographic and specimen identifiers. Statistical analysis of these characters was carried out on a Macintosh IIsi microcomputer using SPSS for the Macintosh (SPSS, Inc., 1990).

Field observations and collection of leaf tissue for isozyme electrophoresis were carried out in the following localities (population acronym and number of individuals in parentheses): Colorado: Routt County, Routt National Forest, Mount Zirkel Wilderness Area, Three Island Lake (TIL, 20 individuals); Oregon: Marion Co., Mount Hood National Forest, Breitenbush Lake Road (BLR, 40 individuals); Washington: Clallam County, Olympic National Park, Hurricane Hill trailhead (ONP, 20 individuals); and British Columbia: Vancouver Island, Grouse Mountain Resort (GMR, 40 individuals). Voucher specimens were deposited at COLO and KANU.

Leaf samples in individual plastic bags were transported to the laboratory on wet ice, ground in buffer as described by Mitton et al. (1979), and stored at -80°C in microfuge tubes until use.

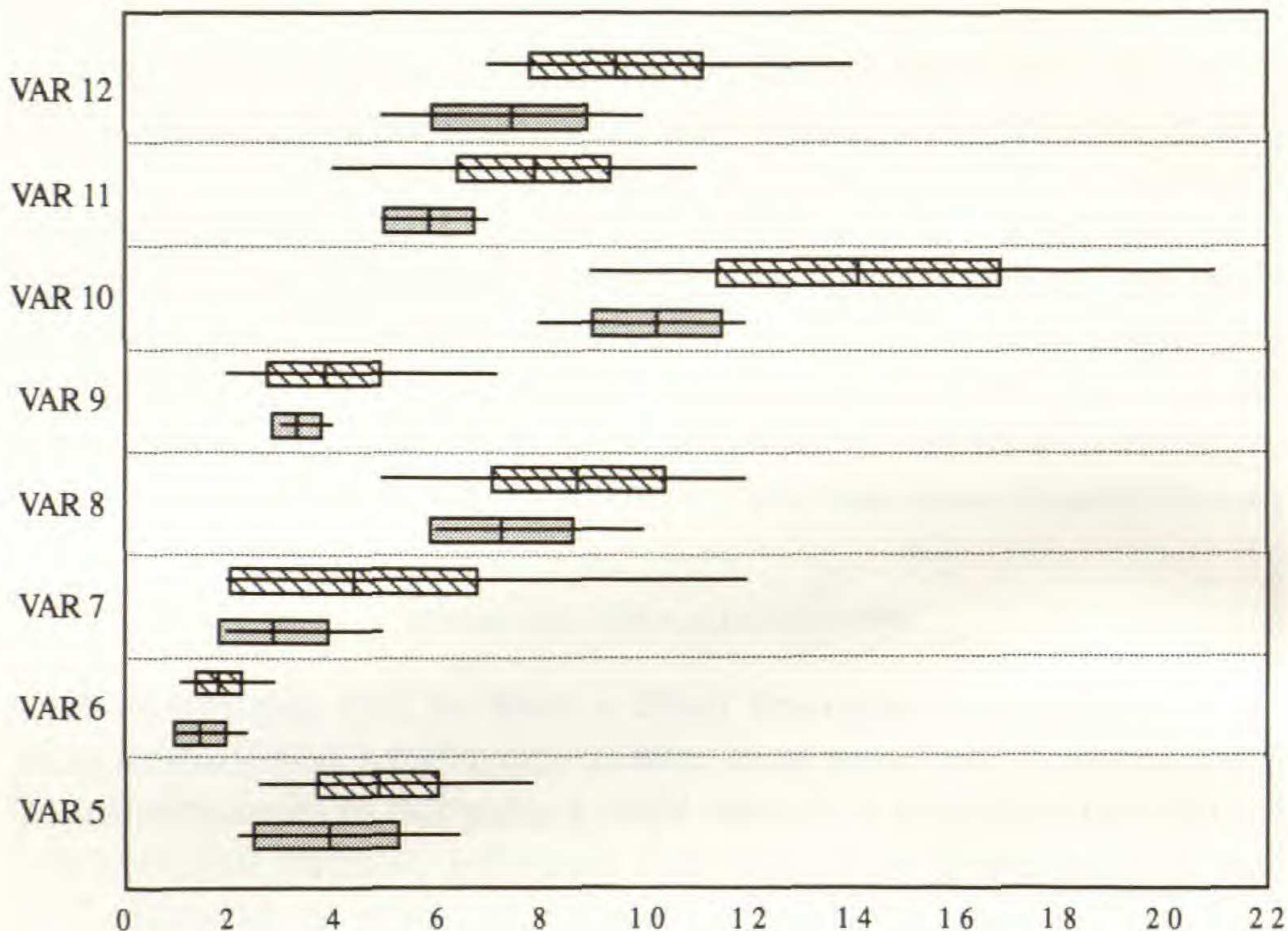


Figure 2. Graphic representation of the statistical analysis of eight meristic characters (see Materials and Methods for explanation) of *Rhododendron albiflorum* specimens from the Pacific Northwest ($n = 57$) and Colorado ($n = 7$). Numbers on x-axis are cm for variables 5-6 and mm for variables 7-12. Thin line = range, boxed area = mean \pm one standard deviation (hatched fill = Pacific Northwest, solid fill = Colorado), vertical bar = mean. Note that while the means for some characters (e.g., petal length, VAR10, and petal width, VAR11) are significantly different (Table 1), the range for the Colorado populations is included within (VAR11) or greatly overlaps (VAR10) that of the Pacific Northwest populations, reducing their usefulness as taxonomic characters.

Starch gels were prepared according to protocols of Soltis et al. (1983). Interpretable banding patterns were obtained from the enzymes glucose-6-phosphate isomerase, phosphoglucomutase, leucine amino peptidase, aspartate amino transferase and triose-phosphate isomerase when run in buffer system 6 or a modification of buffer system 8 (Haufler, 1985). The clearest banding patterns for isocitric dehydrogenase, shikimate dehydrogenase, malate dehydrogenase, and 6-phosphogluconate dehydrogenase were obtained using buffer system 11 of Soltis et al. (1983) or a pH 7.5 modification of the morpholine system of Odrzykoski and Gottlieb (1984). Data from banding patterns were analyzed using the program BIOSYS-1 (Swofford and Selander, 1981) on a Zenith 159 microcomputer.

Table 1. Comparison of means and standard deviations (SD) for eight measured characteristics (VAR5 through VAR12, as explained in text) of Colorado and Pacific Northwest (Oregon, Washington, British Columbia, Alberta, Montana and Idaho) specimens of *Rhododendron albiflorum*. Measurements were taken from herbarium sheets; flowers were rehydrated in Tween™.

Character	Mean (SD)		P
	Colorado (n = 7)	Pacific Northwest (n = 51)	
VAR5 Leaf L (cm)	3.8714 (1.378)	4.8314 (1.150)	<.05
VAR6 Leaf W (cm)	1.4571 (0.469)	1.8196 (0.444)	<.05
VAR7 Flowers/cluster	2.8571 (1.069)	4.4118 (2.376)	NS
VAR8 Sepal L (mm)	7.2857 (1.380)	8.7549 (1.650)	<.05
VAR9 Sepal W (mm)	3.2857 (0.488)	3.8333 (1.109)	NS
VAR10 Petal L (mm)	10.2857 (1.254)	14.1373 (2.764)	.001
VAR11 Petal W (mm)	5.8571 (0.900)	7.8725 (1.466)	.001
VAR12 Stamen L (mm)	7.4286 (1.512)	9.4706 (1.669)	<.01

RESULTS AND DISCUSSION

The original descriptions provided by Hooker (1838) and Nelson (1913) differ in language and length, but little in content. In our examination of the plants, we found no qualitative characters, such as color or shape of corollas or shape of leaves, that could be used to distinguish two different species. Likewise, pollen from plants of the two areas was identical. Although Colorado plants tend to be shorter (usually <1 m) than Pacific Northwest plants (1–2 m), and to have smaller leaves and fewer and smaller flowers, these differences may be attributable to phenotypic plasticity in response to the severity of the habitats in which they grow. Elevational range for the Pacific Northwest populations is 1160–2700 m above sea level and weather patterns have an oceanic influence, while for the Colorado populations, the elevational range is 3000–3500 m above sea level and the climate is distinctly continental.

Analysis of the quantitative characters indicated that the means of measurements of leaf length and width, sepal length, petal length and width, and stamen length of the Colorado populations (Table 1) were significantly smaller than the means of the same measurements for the Pacific Northwest populations. However, correlations (Table 2) among the characters that have significantly different means (Table 1) reduce the taxonomic value of those

Table 2. Correlation coefficients among variables measured on specimens of *Rhododendron albiflorum* ($n = 58$). Variables 1-4 were specimen and locality identification codes. VAR5 = leaf length, VAR6 = leaf width, VAR7 = number of flowers per axillary cluster, VAR8 = sepal length, VAR9 = sepal width, VAR10 = petal length, VAR11 = petal width, VAR12 = stamen length.

	VAR5	VAR6	VAR7	VAR8	VAR9	VAR10	VAR11	VAR12
VAR5	1.0000	.8284**	.2225	.2414	.1053	.0618	.0831	.1632
VAR6		1.0000	.2721*	.2817*	.1628	.1084	.0748	.2404
VAR7			1.0000	.1220	-.1033	.0920	-.3019*	.0862
VAR8				1.0000	.3881**	.5345**	.4620**	.4026*
VAR9					1.0000	.1874	.2544	.2075
VAR10						1.0000	.6019**	.6156*
VAR11							1.0000	.4794*
VAR12								1.0000

* Significant at the .05 level.

** Significant at the .01 level.

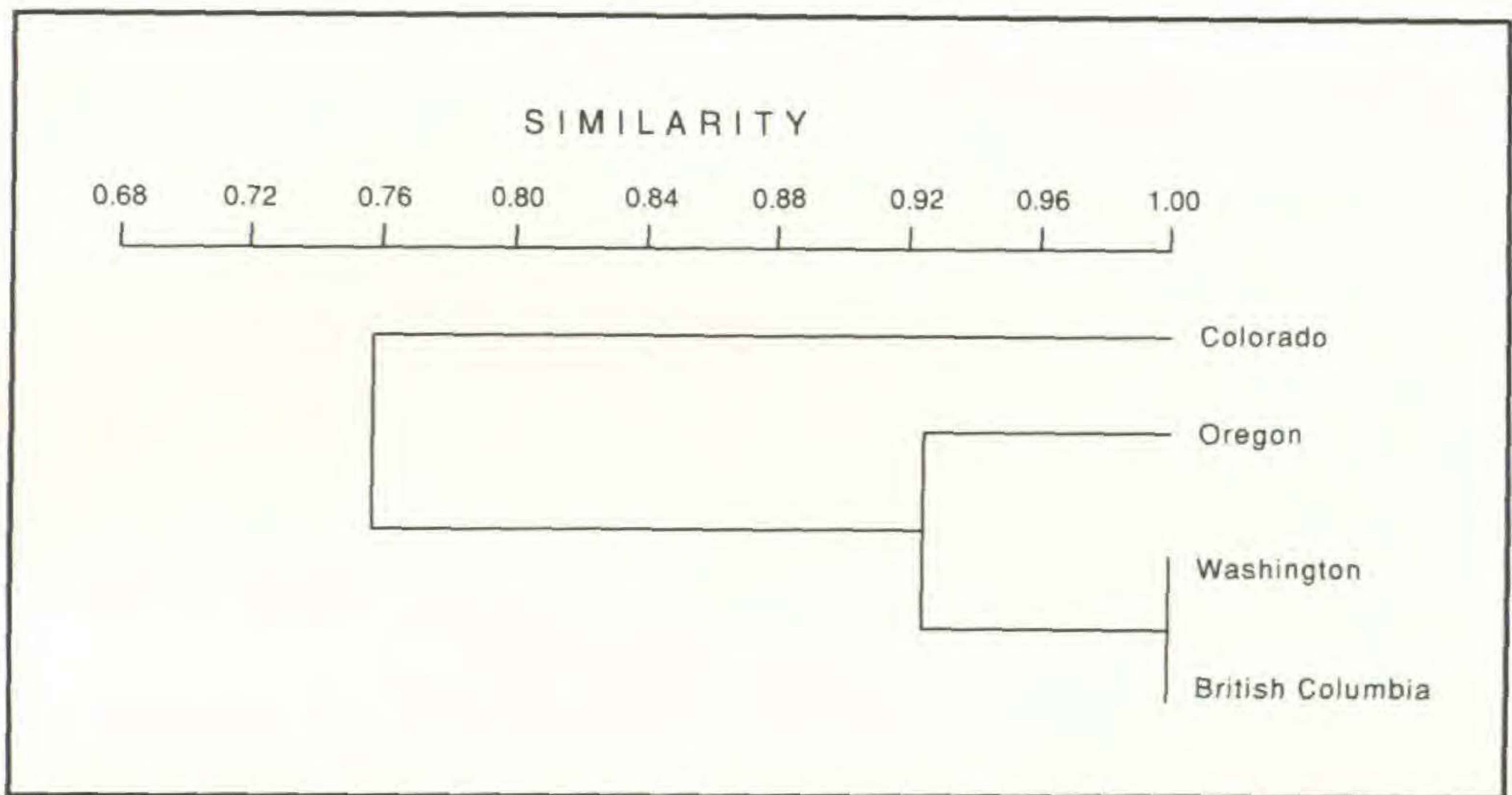


Figure 3. Dendrogram produced by cluster analysis calculated by the unweighted pair group method (Swofford and Selander, 1981) based on unbiased genetic identity (Nei, 1978).

differences. For example, it is reasonable for a shorter petal also to be narrower (VAR10 is correlated at the .01 level with VAR11) or for shorter leaves also to be narrower (the correlation between VAR5 and VAR6 is significant at the .01 level). Even though a discriminant function analysis of the Colorado populations versus the Pacific Northwest populations indicated that petal length and width, and stamen length, accounted for most of the distinction between the groups, the ranges of measurements of these characters for the Colorado populations are either completely within or significantly overlapping with those for the Pacific Northwest populations (Figure 2). Finally, a cluster analysis (not illustrated) performed on all 58 specimens failed to group the 7 Colorado specimens; rather, these populations were scattered among those from Montana, Alberta and Washington. Thus, we conclude that the morphological evidence does not support the recognition of the Colorado plants as a species distinct from that of the Pacific Northwest.

Results from isozymic analysis showed the Colorado populations to be distinct but failed to provide compelling evidence for recognizing these plants at the specific level. As summarized in Table 3 and Figure 3, genetic identity values among the Pacific Northwest populations were consistently above .90 while those between the Colorado and Pacific Northwest populations ranged from .729 to .722. Given that the average genetic identity (Nei,

Table 3. Matrix of similarity and distance coefficients (Nei, 1978) based on data from electrophoretic analysis of 10 enzyme systems. Population acronyms are explained in Materials and Methods. Above diagonal: unbiased genetic distance; below diagonal: unbiased genetic identity.

Population	1	2	3	4
1 TIL (Colorado)	—	.273	.258	.316
2 BLR (Oregon)	.761	—	.118	.090
3 ONP (Washington)	.772	.913	—	.000
4 GMR (British Columbia)	.729	.939	1.000	—

1978) among congeneric angiosperm species is .67 (Crawford, 1983), our results might be used to suggest that the Colorado populations should be elevated to specific status. However, it is probable that the Colorado populations represent glacial relics (Davis, 1983) that have been isolated for sufficient time to diverge at the isozyme level, but there has been no corresponding morphological divergence except in size.

Thus, the combined data suggest that the most reasonable taxonomic treatment of the two sets of populations is the recognition of two varieties of a single species. The Pacific Northwest plants are referred to *R. albiflorum* Hook var. *albiflorum*.

Rhododendron albiflorum Hook. var. **warrenii** (A. Nelson) M. A. Lane, *comb. & stat. nov.*

Azaleastrum warrenii A. Nelson. TYPE: Colorado, Jackson Co., lower slope of Mt. Zirkel, 9275 ft., 14 Jul 1911, *E. R. Warren 16* [HOLOTYPE: RM!].

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LITERATURE CITED

CRAWFORD, D. J. 1983. Phylogenetic and systematic inferences from electrophoretic studies, pp. 257–287. *In*: S. O. Tanksley and T. J. Orton, Eds.,

- Isozymes in Plant Genetics and Breeding, Part A. Elsevier Press, Amsterdam, The Netherlands.
- DAVIS, M. E. 1983. Quaternary history of deciduous forests of eastern North America and Europe. *Ann. Missouri Bot. Gard.* 70: 550–563.
- HARRINGTON, H. D. 1954. *Manual of the Plants of Colorado*. Sage Books, Denver, CO.
- HAUFLER, C. H. 1985. Enzyme variability and modes of evolution in *Bommeria* (Pteridaceae). *Syst. Bot.* 10: 92–104.
- HITCHCOCK, C. L. AND A. CRONQUIST. 1973. *Flora of the Pacific Northwest*. University of Washington Press, Seattle, WA.
- HOOVER, J. D. 1838. *Flora Boreali Americana*, Vol. 2. H. G. Bohn, London. p. 43.
- MCKELVEY, S. D. 1956. Botanical Exploration of the Trans-Mississippi West 1790–1850. p. 486. The Arnold Arboretum of Harvard University, Jamaica Plain, MA.
- MITTON, J. B., Y. B. LINHART, K. B. STURGEON AND J. L. HAMRICK. 1979. Allozyme polymorphisms detected in mature needle tissue of ponderosa pine. *J. Heredity* 70: 86–89.
- NEI, M. 1978. Estimates of average heterozygosity and genetic distance from a small number of individuals. *Genetics* 89: 583–590.
- NELSON, A. 1913. Contributions from the Rocky Mountain Herbarium XIII. *Bot. Gaz. (Crawfordsville)* 56: 63–68.
- ODRZYKOSKI, I. J. AND L. D. GOTTLIEB. 1984. Duplications of genes encoding 6-phosphogluconate dehydrogenase in *Clarkia* (Onagraceae) and their phylogenetic implications. *Syst. Bot.* 9: 479–489.
- PALSER, B. F., W. R. PHILIPSON AND M. N. PHILIPSON. 1991. Characteristics of ovary, ovule, and mature megagametophyte in *Rhododendron* L. and their taxonomic significance. *Bot. J. Linn. Soc.* 105: 289–390.
- PECK, M. G. 1961. *A Manual of the Higher Plants of Oregon*. Oregon State University Press, Portland, OR.
- PHILIPSON, W. R. AND M. N. PHILIPSON. 1986. A revision of *Rhododendron*. III. Subgenera *Azaleastrum*, *Mumeazalea*, *Candidastrum*, and *Therorhodium*. *Notes Roy. Bot. Gard. Edinburgh* 44: 1–23.
- RYDBERG, P. A. 1900. Rocky Mountain plants. *Mem. New York Bot. Gard.* 1: 297.
- SOLTIS, D. E., C. H. HAUFLER, D. C. DARROW AND G. J. GASTONY. 1983. Starch gel electrophoresis of ferns: a compilation of grinding buffers, gel and electrode buffers and staining schedules. *Amer. Fern J.* 73: 9–27.
- SPSS, INC. 1990. *SPSS for the Macintosh, Version 4.0*. SPSS Inc., Chicago, IL.
- SWOFFORD, D. AND R. B. SELANDER. 1981. BIOSYS-1: A Computer Program for the Analysis of Allelic Variation in Genetics. University of Illinois, Urbana-Champaign, IL.
- WEBER, W. A. 1990. *Colorado Flora: Eastern Slope*. University Press of Colorado, Niwot, CO.
- WEBER, W. A. AND R. C. WITTMANN. 1992. *Catalog of the Colorado Flora: A Biodiversity Baseline*. University Press of Colorado, Niwot, CO.

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